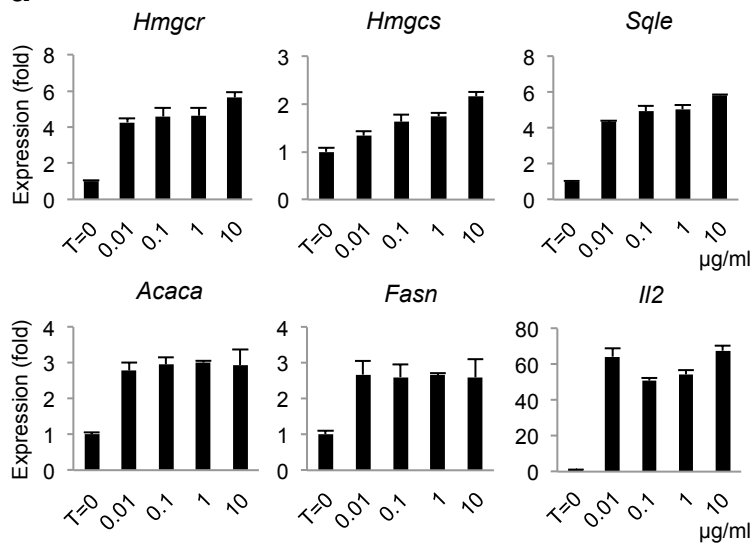
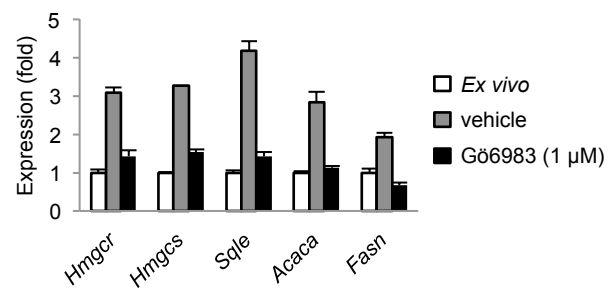


# Supplementary figure 1

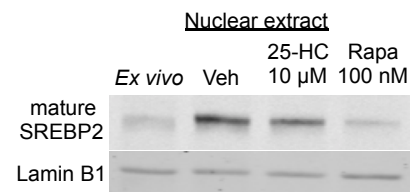
**a**



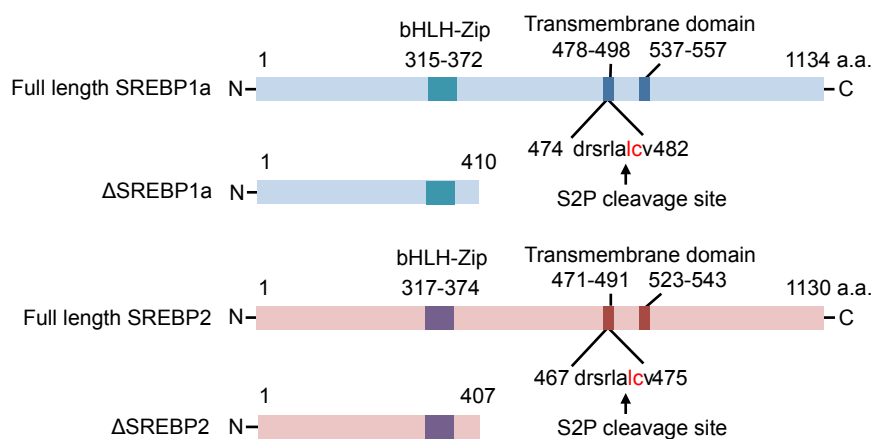
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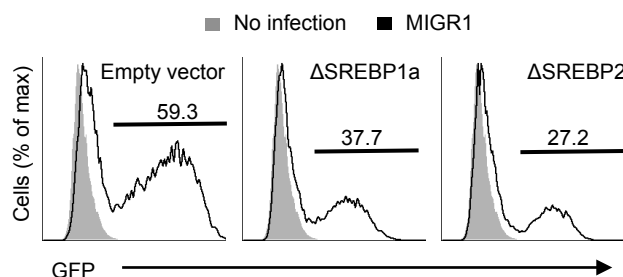
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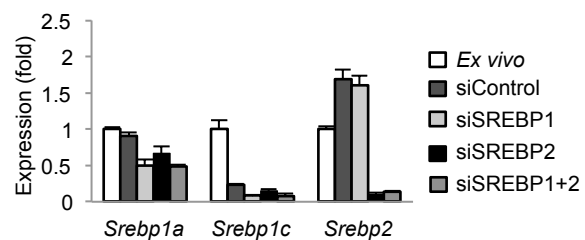
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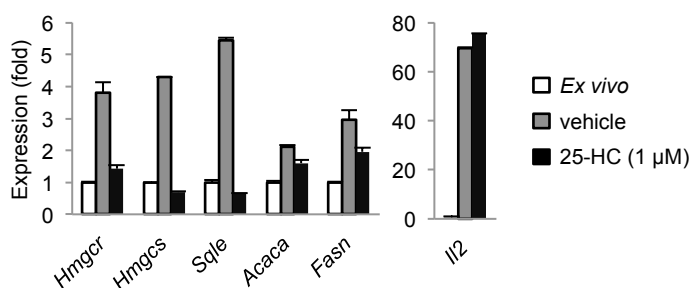
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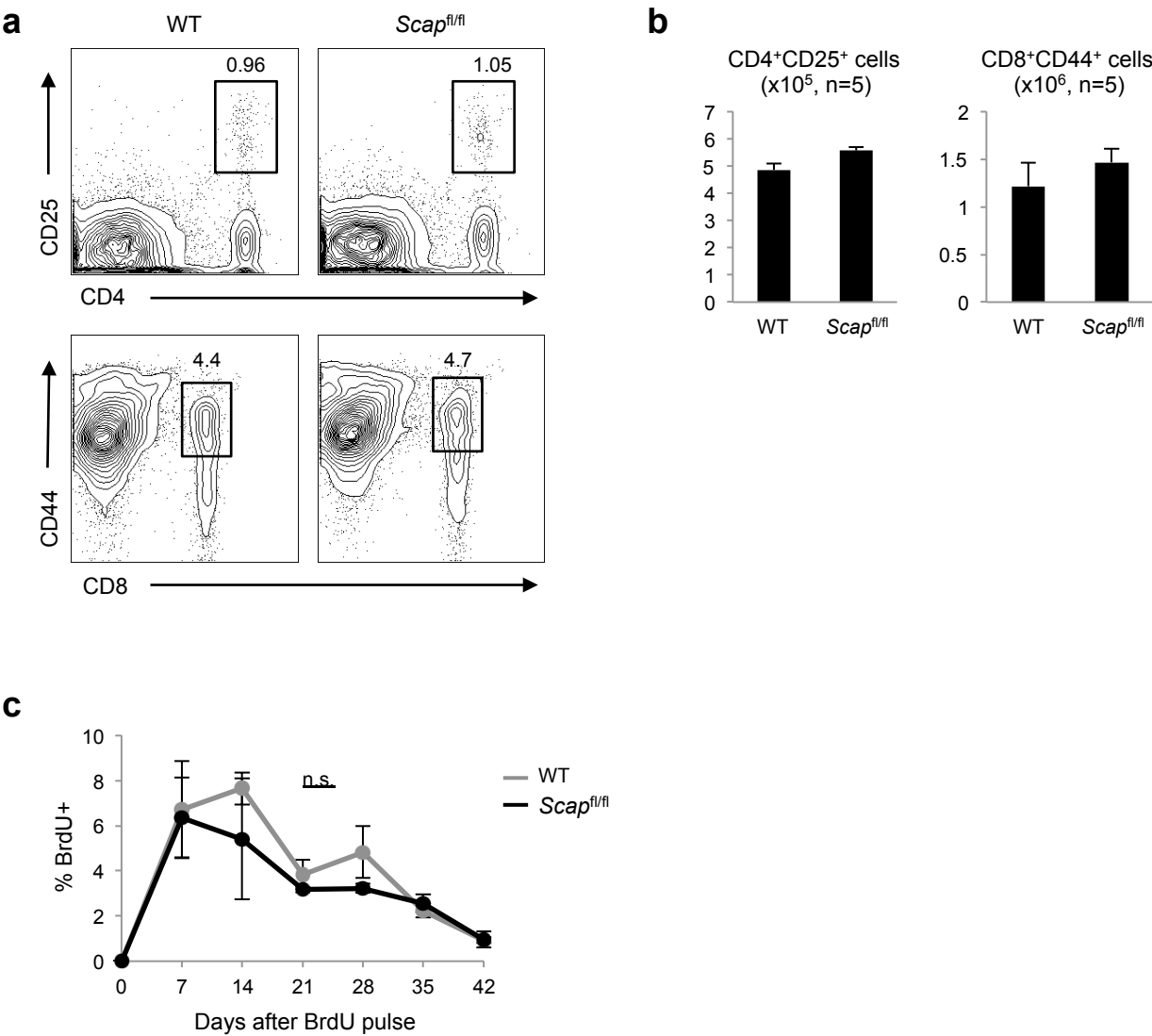
**f**



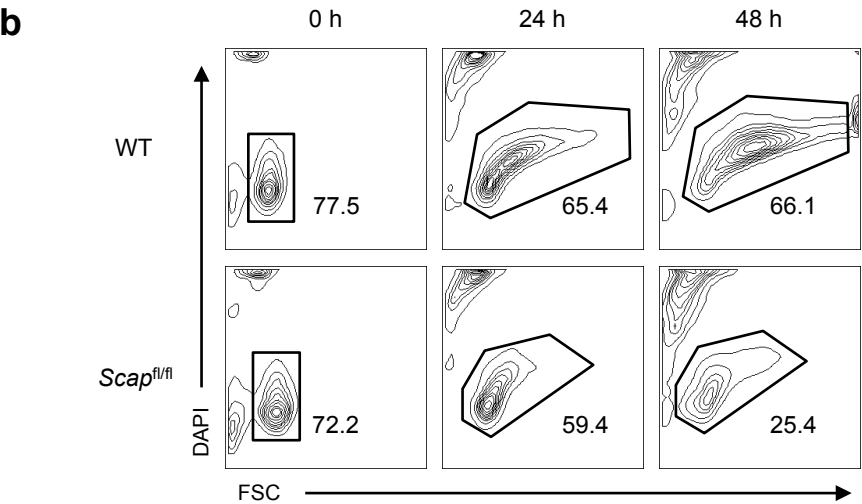
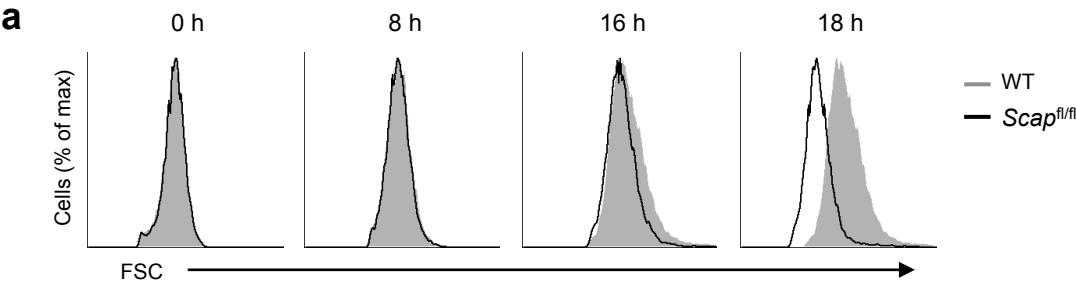
**g**



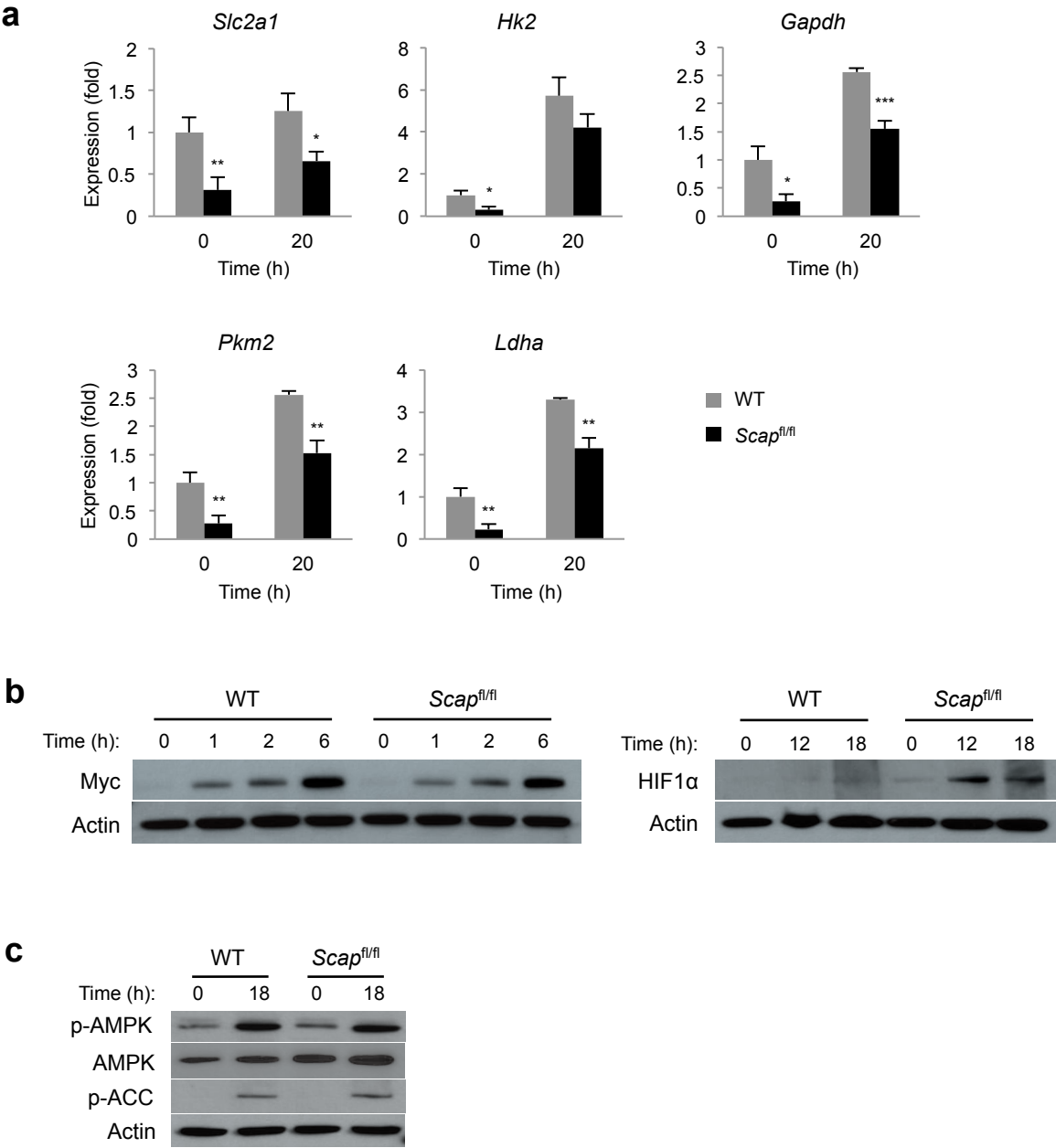
Supplementary Figure 2



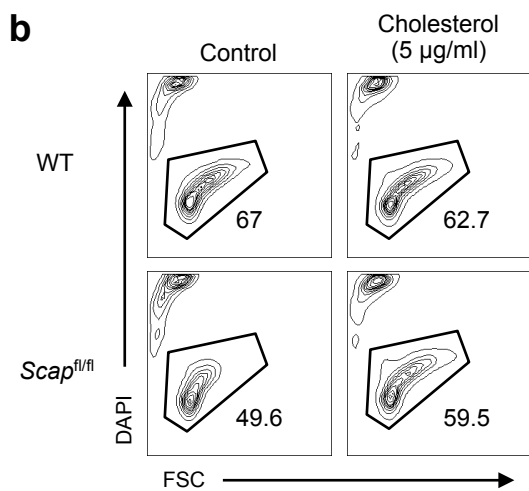
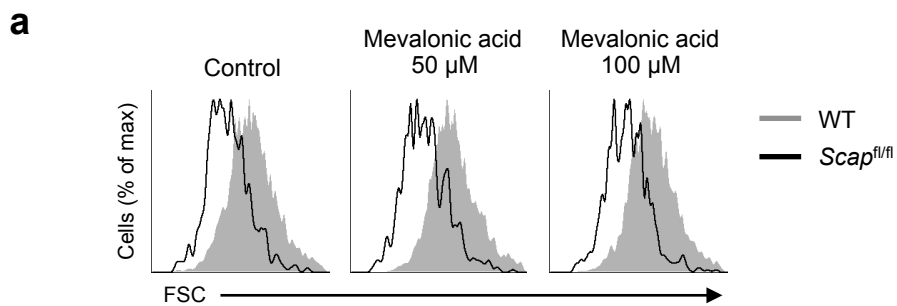
Supplementary Figure 3



Supplementary Figure 4



## Supplementary Figure 5



Supplemental Table 1  
Genes downregulated in activated SCAP-deficient T cells

1421684_at	Ctsll3	Cathepsin L-like 3
1417143_at	Lpar1	Lysophosphatidic acid receptor 1
1420990_at	Chd1	Chromodomain helicase DNA binding protein 1
1436343_at	Chd4	Chromodomain helicase DNA binding protein 4
1443904_at	#N/A	#N/A
1448794_s_at	Dnajc2	DnaJ (Hsp40) homolog, subfamily C, member 2
1458847_at	0	Transcribed locus
1417775_at	Polr1a	Polymerase (RNA) I polypeptide A
1419176_at	Vps37a	Vacuolar protein sorting 37A (yeast)
1426483_at	Prkrir	Protein-kinase, interferon-inducible double stranded RNA dependent inhibitor, repressor of (P58 repressor)
1433613_at	Pank3	Pantothenate kinase 3
1421894_a_at	Tpp2	Tripeptidyl peptidase II
1415807_s_at	Srsf2	Serine/arginine-rich splicing factor 2
1418964_at	Pigm	Phosphatidylinositol glycan anchor biosynthesis, class M
1417871_at	Hsd17b7	Hydroxysteroid (17-beta) dehydrogenase 7
1423143_at	Gtpbp4	GTP binding protein 4
1426853_at	0	Transcribed locus, moderately similar to NP_001025862.1 protein SET [Gallus gallus]
1433445_x_at	Hmgcs1	3-hydroxy-3-methylglutaryl-Coenzyme A synthase 1
1422533_at	Cyp51	Cytochrome P450, family 51
1450646_at	Cyp51	Cytochrome P450, family 51
1433443_a_at	Hmgcs1	3-hydroxy-3-methylglutaryl-Coenzyme A synthase 1
1450010_at	Hsd17b12	Hydroxysteroid (17-beta) dehydrogenase 12
1433444_at	Hmgcs1	3-hydroxy-3-methylglutaryl-Coenzyme A synthase 1
1448563_at	Phb	Prohibitin
1440346_at	Kdm6b	KDM1 lysine (K)-specific demethylase 6B
1449072_a_at	N6amt2	N-6 adenine-specific DNA methyltransferase 2 (putative)
1450701_a_at	Gtf2h2	General transcription factor II H, polypeptide 2
1426630_at	Exosc2	Exosome component 2
1426831_at	Ahcy1	S-adenosylhomocysteine hydrolase-like 1
1438015_at	Dkc1	Dyskeratosis congenita 1, dyskerin homolog (human)
1422769_at	Syncrip	Synaptotagmin binding, cytoplasmic RNA interacting protein
1444498_at	Gphn	Gephyrin

Supplemental Table 1  
Genes downregulated in activated SCAP-deficient T cells

1459403_at	Ldlr	Low density lipoprotein receptor
1450743_s_at	Syncrip	Synaptotagmin binding, cytoplasmic RNA interacting protein
1428479_at	Nfatc1	Nuclear factor of activated T-cells, cytoplasmic, calcineurin-dependent 1
1433446_at	Hmgcs1	3-hydroxy-3-methylglutaryl-Coenzyme A synthase 1
1450172_at	Pknox1	Pbx/knotted 1 homeobox
1456058_at	Rbm27	RNA binding motif protein 27
1415867_at	Cct4	Chaperonin containing Tcp1, subunit 4 (delta)
1433535_x_at	Cct2	Chaperonin containing Tcp1, subunit 2 (beta)
1434038_at	Dnajc13	DnaJ (Hsp40) homolog, subfamily C, member 13
1444268_at	BC004004	CDNA sequence BC004004
1430980_a_at	0	Transcribed locus, strongly similar to NP_989880.1 eukaryotic initiation factor 4A-II [Gallus gallus]
1418563_at	Serbp1	Serpine1 mRNA binding protein 1
1420157_s_at	Abcf1	ATP-binding cassette, sub-family F (GCN20), member 1
1421914_s_at	Mrpl19	Mitochondrial ribosomal protein L19
1459530_at	#N/A	#N/A
1425113_x_at	#N/A	#N/A
1457248_x_at	#N/A	#N/A
1450846_at	Bzw1	Basic leucine zipper and W2 domains 1
1415993_at	Sqle	Squalene epoxidase
1423078_a_at	Sc4mol	Sterol-C4-methyl oxidase-like
1459315_at	0	Transcribed locus
1415773_at	Ncl	Nucleolin
1423804_a_at	Idi1	Isopentenyl-diphosphate delta isomerase
1430290_at	0	Transcribed locus, strongly similar to NP_997927.1 40S ribosomal protein S15a [Danio rerio]
1427229_at	Hmgcr	3-hydroxy-3-methylglutaryl-Coenzyme A reductase
1450087_a_at	Nolc1	Nucleolar and coiled-body phosphoprotein 1
1431235_at	Matr3	Matrin 3
1451122_at	Idi1	Isopentenyl-diphosphate delta isomerase
1434752_at	0	Transcribed locus, strongly similar to NP_001019.1 40S ribosomal protein S25 [Homo sapiens]

Supplemental Table 1  
Genes downregulated in activated SCAP-deficient T cells

1428870_at	Nolc1	Nucleolar and coiled-body phosphoprotein 1
1450986_at	Nop58	NOP58 ribonucleoprotein homolog (yeast)
1430778_a_at	Nubp1	Nucleotide binding protein 1
1423418_at	Fdps	Farnesyl diphosphate synthetase
1423151_at	Dnajb11	DnaJ (Hsp40) homolog, subfamily B, member 11
1447568_at	Gatc	Glutamyl-tRNA(Gln) amidotransferase, subunit C homolog (bacterial)
1453424_at	#N/A	#N/A
1456610_at	Kdm6b	KDM1 lysine (K)-specific demethylase 6B
1436384_at	0	Transcribed locus, strongly similar to XP_418029.1 PREDICTED: hypothetical protein [Gallus gallus]
1457199_at	0	Transcribed locus
1426596_a_at	Smn1	Survival motor neuron 1
1434630_at	Ankrd28	Ankyrin repeat domain 28
1455330_at	Nol9	Nucleolar protein 9
1435187_at	0	Transcribed locus
1440253_at	Psmd11	Proteasome (prosome, macropain) 26S subunit, non-ATPase, 11
1456081_a_at	Aacs	Acetoacetyl-CoA synthetase
1451345_at	Mtap	Methylthioadenosine phosphorylase
1420067_at	Cd160	CD160 antigen
1435159_at	9430038I01Rik	RIKEN cDNA 9430038I01 gene
1439972_at	Etnk1	Ethanolamine kinase 1
1431239_at	Nono	Non-POU-domain-containing, octamer binding protein
1440513_at	#N/A	#N/A
1437667_a_at	Bach2	BTB and CNC homology 2
1458393_at	#N/A	#N/A
1454664_a_at	Eif5	Eukaryotic translation initiation factor 5
1416372_at	Ptdss1	Phosphatidylserine synthase 1
1447947_at	Zfyve16	Zinc finger, FYVE domain containing 16
1419914_s_at	Nus1	Nuclear undecaprenyl pyrophosphate synthase 1 homolog (S. cerevisiae)
1450132_at	Hivp3	Human immunodeficiency virus type I enhancer binding protein 3
1440996_at	0	Transcribed locus
1449915_at	Zfp202	Zinc finger protein 202
1417442_a_at	Pex3	Peroxisomal biogenesis factor 3



Supplemental Table 1  
Genes downregulated in activated SCAP-deficient T cells

at		
1449732_at	Zscan21	Zinc finger and SCAN domain containing 21
1415911_at	Impact	Imprinted and ancient
1454954_at	Wdr61	WD repeat domain 61
1424250_a_at	Arhgef3	Rho guanine nucleotide exchange factor (GEF) 3
1423041_a_at	Bzw1	Basic leucine zipper and W2 domains 1
1432850_at	0	Transcribed locus
1416244_a_at	Cnbp	Cellular nucleic acid binding protein
1431502_a_at	Exosc3	Exosome component 3
1435099_at	Utp14a	UTP14, U3 small nucleolar ribonucleoprotein, homolog A (yeast)
1439154_at	Nup98	Nucleoporin 98
1426482_at	Prkrir	Protein-kinase, interferon-inducible double stranded RNA dependent inhibitor, repressor of (P58 repressor)
1429710_at	Styx	Serine/threonine/tyrosine interaction protein
1450858_a_at	Ube2d3	Ubiquitin-conjugating enzyme E2D 3 (UBC4/5 homolog, yeast)
1438897_at	Zdhhc3	Zinc finger, DHHC domain containing 3
1441578_at	Ccdc86	Coiled-coil domain containing 86
1448263_a_at	Cndp2	CNDP dipeptidase 2 (metallopeptidase M20 family)
1438383_x_at	0	Transcribed locus
1436183_at	Zc3hav1	Zinc finger CCCH type, antiviral 1
1445269_at	#N/A	#N/A
1449039_a_at	Hnrpd1	Heterogeneous nuclear ribonucleoprotein D-like
1427923_at	Zmpste24	Zinc metallopeptidase, STE24 homolog (S. cerevisiae)
1442135_at	Ado	2-aminoethanethiol (cysteamine) dioxygenase
1441657_at	0	Transcribed locus, weakly similar to XP_002729512.1 PREDICTED: hypothetical protein [Rattus norvegicus]
1435953_at	Btaf1	BTA1 RNA polymerase II, B-TFIID transcription factor-associated, (Mot1 homolog, S. cerevisiae)
1451087_at	Wdr36	WD repeat domain 36
1454923_at	Iws1	IWS1 homolog (S. cerevisiae)
1439867_at	0	Transcribed locus
1455517_at	Rbm4	RNA binding motif protein 4
1420129_s_	D10Wsu52e	DNA segment, Chr 10, Wayne State University 52, expressed

Supplemental Table 1  
Genes downregulated in activated SCAP-deficient T cells

at		
1435302_at	Taf4b	TAF4B RNA polymerase II, TATA box binding protein (TBP)-associated factor
1434884_at	Mtdh	Metadherin
1447270_at	#N/A	#N/A
1436007_a_at	Thumpd1	THUMP domain containing 1
1421969_a_at	Faah	Fatty acid amide hydrolase
1417766_at	Cyb5b	Cytochrome b5 type B
1426744_at	Srebf2	Sterol regulatory element binding factor 2
1440841_at	0	Transcribed locus
1421821_at	Ldlr	Low density lipoprotein receptor
1437878_s_at	Ttc14	Tetratricopeptide repeat domain 14
1451895_a_at	Dhcr24	24-dehydrocholesterol reductase
1441233_at	#N/A	#N/A
1448463_at	4933434E20Rik	RIKEN cDNA 4933434E20 gene
1448865_at	Hsd17b7	Hydroxysteroid (17-beta) dehydrogenase 7
1420852_a_at	B3gnt2	UDP-GlcNAc:betaGal beta-1,3-N-acetylglucosaminyltransferase 2
1432867_at	#N/A	#N/A
1442408_at	Sulf2	Sulfatase 2
1418844_at	Alg9	Asparagine-linked glycosylation 9 homolog (yeast, alpha 1,2 mannosyltransferase)
1442992_at	#N/A	#N/A
1445748_at	#N/A	#N/A
1426074_at	0	Hypothetical protein LOC635138, mRNA (cDNA clone MGC:28021 IMAGE:3660814)
1431947_at	Ldlr	Low density lipoprotein receptor
1447960_at	0	Transcribed locus
1451457_at	Sc5d	Sterol-C5-desaturase (fungal ERG3, delta-5-desaturase) homolog ( <i>S. cerevisiae</i> )
1454671_at	Insig1	Insulin induced gene 1
1438294_at	Atxn1	Ataxin 1
1456589_x_at	#N/A	#N/A

Supplemental Table 1  
Genes upregulated in activated SCAP-deficient T cells

1416392_a_at	Atp6v0c	ATPase, H <sup>+</sup> transporting, lysosomal V0 subunit C
1430163_at	Rab43	RAB43, member RAS oncogene family
1430252_at	0	Transcribed locus
1435588_at	Wdfy1	WD repeat and FYVE domain containing 1
1416834_x_at	Ndufb2	NADH dehydrogenase (ubiquinone) 1 beta subcomplex, 2
1422536_at	Tnni3	Troponin I, cardiac 3
1420397_a_at	Spen	SPEN homolog, transcriptional regulator (Drosophila)
1437358_at	Wdfy1	WD repeat and FYVE domain containing 1
1453207_at	0	Transcribed locus, strongly similar to XP_001069626.1 PREDICTED: similar to CG14483-PA [Rattus norvegicus]
1416083_at	Zfand5	Zinc finger, AN1-type domain 5
1416512_at	Nubp2	Nucleotide binding protein 2
1422488_at	Nxt1	NTF2-related export protein 1
1421947_at	Gng12	Guanine nucleotide binding protein (G protein), gamma 12
1455412_at	Sfmbt1	Scm-like with four mbt domains 1
1449295_at	Sap30b p	SAP30 binding protein
1449523_at	Bcl7c	B-cell CLL/lymphoma 7C
1416004_at	Ywhah	Tyrosine 3-monooxygenase/tryptophan 5-monooxygenase activation protein, eta polypeptide
1416082_at	Cep68	Centrosomal protein 68
1434597_at	0	Transcribed locus
1450275_x_at	Ube2f	Ubiquitin-conjugating enzyme E2F (putative)
1460450_at	0	Transcribed locus
1430934_at	Naaladl 2	N-acetylated alpha-linked acidic dipeptidase-like 2
1418011_a_at	0	Transcribed locus
1436869_at	Shh	Sonic hedgehog
1439379_x_at	Prm1	Protamine 1
1417838_at	Ssty2	Spermiogenesis specific transcript on the Y 2
1420310_at	#N/A	#N/A
1416666_at	Serpine 2	Serine (or cysteine) peptidase inhibitor, clade E, member 2
1447044_at	#N/A	#N/A
1429169_at	Rbm3	RNA binding motif protein 3
1457507_at	0	Transcribed locus
1449360_at	Csf2rb2	Colony stimulating factor 2 receptor, beta 2, low-affinity (granulocyte-macrophage)
1418092_s_at	Trip10	Thyroid hormone receptor interactor 10
1438050_x_at	#N/A	#N/A
1459250_at	0	Transcribed locus, strongly similar to NP_536703.1 teashirt zinc

Supplemental Table 1  
Genes upregulated in activated SCAP-deficient T cells

		finger family member 2 [Mus musculus]	
1455394_at	Pias4	Protein inhibitor of activated STAT 4	
1442163_at	#N/A	#N/A	
1447393_at	#N/A	#N/A	
1422415_at	Ang2	Angiogenin, ribonuclease A family, member 2	
1430002_at	lqcf1	IQ motif containing F1	
1456901_at	Adamts 20	A disintegrin-like and metallopeptidase (reprolysin type) with thrombospondin type 1 motif, 20	
1423256_a_at	0	Transcribed locus	
1438669_at	Dcaf12	DDB1 and CUL4 associated factor 12	
1443609_s_at	Syvn1	Synovial apoptosis inhibitor 1, synoviolin	
1434861_at	0	Transcribed locus	
1428939_s_at	Gnaq	Guanine nucleotide binding protein, alpha q polypeptide	
1442171_at	0	Transcribed locus	
1441351_at	#N/A	#N/A	
1442361_at	#N/A	#N/A	
1452124_at	Ank3	Ankyrin 3, epithelial	

Supplemental Table 1  
Genes downregulated in quiescent SCAP-deficient T cells

1416568_a_at	Acin1	Apoptotic chromatin condensation inducer 1	
1416801_at	Trpm7	Transient receptor potential cation channel, subfamily M, member 7	
1428127_at	Atxn7l3b	Ataxin 7-like 3B	
1455151_at	Akap9	A kinase (PRKA) anchor protein (yotiao) 9	
1438476_a_at	Chd4	Chromodomain helicase DNA binding protein 4	
1439180_at	Ino80d	INO80 complex subunit D	
1424179_at	Plekhj1	Pleckstrin homology domain containing, family J member 1	
1440314_at	0	Transcribed locus	
1428378_at	Zc3hav1	Zinc finger CCCH type, antiviral 1	
1453240_a_at	Gcap14	Granule cell antiserum positive 14	
1439033_at	Zcchc7	Zinc finger, CCHC domain containing 7	
1443878_at	Rapgef6	Rap guanine nucleotide exchange factor (GEF) 6	
1458710_at	0	Transcribed locus	
1423460_at	Gigyf1	GRB10 interacting GYF protein 1	
1438736_at	Thoc2	THO complex 2	
1439203_at	#N/A	#N/A	
1451730_at	Zfp62	Zinc finger protein 62	
1454823_at	Wdr37	WD repeat domain 37	
1426690_a_at	Srebf1	Sterol regulatory element binding transcription factor 1	
1419273_at	C80913	Expressed sequence C80913	
1441274_at	#N/A	#N/A	
1452030_a_at	Hnrnpr	Heterogeneous nuclear ribonucleoprotein R	
1429545_at	Ube2i	Ubiquitin-conjugating enzyme E2I	
1427992_a_at	Rab12	RAB12, member RAS oncogene family	
1459907_a_at	Whsc1l1	Wolf-Hirschhorn syndrome candidate 1-like 1 (human)	
1435002_at	Zfp507	Zinc finger protein 507	
1453263_at	Naa35	N(alpha)-acetyltransferase 35, NatC auxiliary subunit	
1437544_at	Fubp1	Far upstream element (FUSE) binding protein 1	
1460279_a_at	Gtf2i	General transcription factor II I	
1458512_at	Tle3	Transducin-like enhancer of split 3, homolog of Drosophila E(spl)	
1458452_at	Ankrd11	Ankyrin repeat domain 11	
1427254_at	Zfp445	Zinc finger protein 445	
1429432_at	Bat2l2	HLA-B associated transcript 2-like 2	
1444004_at	Thoc2	THO complex 2	
1436775_a_at	Ankrd17	Ankyrin repeat domain 17	
1445499_at	Zc3h13	Zinc finger CCCH type containing 13	
1434393_at	Usp34	Ubiquitin specific peptidase 34	
1450071_at	Ash1l	Ash1 (absent, small, or homeotic)-like (Drosophila)	
1438556_a_at	#N/A	#N/A	

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Supplemental Table 1  
Genes downregulated in quiescent SCAP-deficient T cells

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1459283_at	#N/A	#N/A		
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**Supplementary Figure 1. The lipogenic program of activated CD8<sup>+</sup> T cells is**

**SREBP-dependent.** (a) Real-time PCR analysis of the lipogenic program from purified Pmel TCR transgenic CD8<sup>+</sup> T cells activated with increasing dose of agonist gp100 peptide (0-10 µg/ml). (b) Real-time PCR analysis of quiescent and activated C57BL/6 T cells. Cultures were pretreated with PKC inhibitor Gö6983 (1 µM) for 30 min or vehicle as indicated prior to activation. (c) Immunoblot for mature SREBP2 from nuclear extract and full length SREBP2 from whole cell lysate of C57BL/6 splenic T cells. Nuclear or whole cell extracts were collected from purified CD8<sup>+</sup> T cells. In some cases cells were pretreated with 25-HC (10 µM), rapamycin (100 nM) or vehicle for 30 min, and then activated with PMA/iono for 4-24 h as indicated. (d) Schematic of full length murine SREBP domain structure and truncated forms of SREBP (designated ΔSREBP1a or 2) used for transduction of activated T cells. a.a: amino acid. (e) Efficiency of retroviral transduction in purified C57BL/6 T cells 24 h after infection as shown by GFP expression. (f) Real-time PCR analysis of quiescent or 6 h activated T cells transfected with siSREBP1 alone, siSREBP2 alone, siSREBP1 and siSREBP2 or non-target control (siControl). (g) Real-time PCR analysis of lipogenic genes and IL-2 in quiescent and 6 h activated C57BL/6 splenocytes treated with 25-hydroxycholesterol (1 µM 25-HC) or vehicle. qPCR data are presented as the mean of triplicates with standard deviation. All data is representative of at least three independent experiments.

**Supplementary Figure 2. Conditional ablation of the SREBP chaperone protein**

**SCAP does not perturb peripheral T cell activation phenotype or turn over. (a,b)**

Flow cytometric analysis (a) and absolute cell number (b) of CD4<sup>+</sup>CD25<sup>+</sup> or CD8<sup>+</sup>CD44<sup>+</sup>

splenocytes from 5 week old WT and *Scap*<sup>fl/fl</sup> mice. Frequency of gated cells indicated in FACS plots. (c) Percentage of BrdU-positive T cells in total peripheral T cells collected from blood. 5-week-old WT and *Scap*<sup>fl/fl</sup> mice were injected with a single dose of BrdU i.p., then bled on indicated days. Curves were assessed by 2-way ANOVA and determined to be no different (p=0.2).

**Supplementary Figure 3. Conditional ablation of the SREBP chaperone protein**

**SCAP does not alters apoptosis of activated CD8<sup>+</sup> T cells but perturbs late survival.**

(a) Forward scatter histograms of WT and *Scap*<sup>fl/fl</sup> CD8<sup>+</sup> T cells activated with anti-CD3-28 for indicated time. (b) Frequency of DAPI-negative (live) WT and *Scap*<sup>fl/fl</sup> CD8<sup>+</sup> T cells activated with anti-CD3-28 for indicated time. FACS plots are representative of at least two experiments.

**Supplementary Figure 4. Glucose and glycolysis is impaired in activated *Scap*<sup>fl/fl</sup>**

**CD8<sup>+</sup> T cells.** (a) RT-PCR analysis of quiescent or activated CD8<sup>+</sup> T cells for indicated time. Data are mean of triplicates with standard deviation, and representative of three experiments. *Slc2a1*: solute carrier family 2 (facilitated glucose transporter), member1 (Glut1), *Hk2*: hexokinase2, *Gapdh*: glyceraldehyde-3-phosphate dehydrogenase, *Pkm2*: pyruvate kinase, muscle, *Ldha*: lactate dehydrogenase A. (b,c) Immunoblots for Myc, HIF, phospho and total AMPK, and phospho ACC from whole cell lysate of quiescent or activated CD8<sup>+</sup> T cells with anti-CD3-28 for indicated time. Data are representative of at least two experiments. \*\*p<0.01, \*\*\*p<0.001.



**Supplementary Figure 5. Cholesterol but not mevalonic acid rescues *Scap*<sup>fl/fl</sup> CD8<sup>+</sup> T cell growth and proliferation.** (a) Forward scatter histograms of WT and *Scap*<sup>fl/fl</sup> CD8<sup>+</sup> T cells activated with anti-CD3-28 for 40 h with or without mevalonic acid at indicated concentration. (b) Frequency of DAPI-negative (live) WT and *Scap*<sup>fl/fl</sup> CD8<sup>+</sup> T cells activated with anti-CD3-28 for 24 h with or without 5 µg/ml of MβCD-cholesterol in 10% FBS supplemented media.

**Supplementary Table. List of Genes differentially expressed between WT and *Scap*<sup>fl/fl</sup> in quiescent and activated CD8<sup>+</sup> T cells.** List of genes differentially upregulated in control CD8<sup>+</sup> T cells when compared to *Scap*-deficient T cells 6 h after activation with anti-CD3-28. Genes upregulated in activated *Scap*-deficient CD8<sup>+</sup> T cells compared to activated control CD8<sup>+</sup> T cells. Genes downregulated in quiescent *Scap*-deficient T cells compared to quiescent control CD8<sup>+</sup> T cells.